

## CLAIMS

- 1 – A differential culture medium for ~~the~~ enumeration of food and beverage contaminant yeasts of the *Dekkera* and *Brettanomyces* genera, characterized in  
5 ~~that it comprises a nutrient base, by containing~~ ethanol as the only energy source,  
~~that in combination with~~ *p*-cumaric acid, an acid-base indicator with turning points  
in the acid range, an inhibitor-antibiotic for sensitive yeasts to inhibit the growth of  
some yeast species, and optionally a bacteria growth inhibitor and agar-agar,  
10 allows the selective and differential growth of the said yeasts in food and  
beverages products.
- 2 – A culture medium according to claim 1, characterized in that the present amount of ethanol is from 32 to 96 g/L, preferably 48 g/L.
- 15 3 – A culture medium according to claim 1, characterized in that the present amount of *p*-cumaric acid is from 0.05 to 1.0 g/L, preferably 0.1 g/L.
- 4 – A culture medium according to claim 1, characterized in that the nutrient base  
is has a composition of any "Yeast Nitrogen Base" type, in amounts a total amount  
20 ranging from 5 to 10 g/L, preferably 6.7 g/L.
- 5 – A culture medium according to claim 1, characterized in that the inhibitor antibiotic for some of the yeast species is cycloheximide, present in an amount from 0.004 to 0.1 g/L, preferably 0.01 g/L.
- 25 6 – A culture medium according to claim 1, characterized in that the pH indicator  
with a turning point in the acid range is bromocresol green, present in an amount  
of 0.022 g/L.
- 30 7 – A culture medium according to claim 6, characterized in that the medium pH is  
adjusted with a strong acid HCl, or any other acid of similar strength, to a value  
between 4.8 and 6.0, preferably 5.4.

8 – A culture medium according to claim 1, characterized in that it additionally contains a bacteria growth inhibitor, preferably chloramphenicol and/or oxytetracycline, in ~~amounts~~ an amount of 0.1 g/L, to detect and identify yeasts of the *Dekkera* and *Brettanomyces* genera in food and beverage products containing mixed populations of yeasts and bacteria.

9 – A culture medium according to claim 1, characterized in that it contains all the components except agar-agar, to detect and identify yeasts of the *Dekkera* and *Brettanomyces* genera in food and beverage products containing mixed populations of yeasts, bacteria and particularly filamentous fungi.

10 – A differential culture medium for enumeration of food and beverage contaminant yeasts of the *Dekkera* and *Brettanomyces* genera, characterized in that it has the following composition: 5 to 10 g/L, preferably 6.7 g/L, of a "Yeast Nitrogen Base" type medium; 0.004 to 0.1 g/L, preferably 0.01 g/L, of cycloheximide; 0.05 to 1.0 g/L, preferably 0.1 g/L, of *p*-cumaric acid; 0.022 g/L of bromocresol green, or another acid-base indicator with similar turning points; 32 to 96 g/L, preferably 48 g/L, of ethanol; 0.1 g/L of chloramphenicol and/or 0.1 g/L of oxytetracycline, and 20 g/L of agar-agar, the pH of the medium being adjusted between 4.8 and 6.0, preferably 5.4, with ~~a strong acid~~ HCl, or any other acid of similar strenght.

11– A differential culture medium for enumeration of food and beverage contaminant yeasts of the *Dekkera* and *Brettanomyces* genera according to claim 1, characterized by the sterilization of all the components is done by filtration, except for the agar-agar which is sterilized in autoclave; the addition under aseptic conditions to this solution, after agar-agar cooling and before it solidifies, of all the other components of the medium, previously sterilized by filtration; and the dispensing of the medium into Petri dishes so that it solidifies.

~~12 – A process for the detection and/or identification of yeasts of the *Dekkera* and *Brettanomyces* genera, characterized by the use of a differential culture medium for enumeration of food and beverage contaminant yeasts of the *Dekkera* and~~

~~Brettanomyces~~ genera, comprising a nutrient base, ethanol as the only energy source, p-cumalic acid, an acid-base indicator with turning points in the acid range, an inhibitor antibiotic for some of the yeast species, and optionally a bacteria growth inhibitor and agar-agar.

5

13—A process for the detection and/or identification of yeasts of the *Dekkera* and *Brettanomyces* genera according to claim 12, characterized in that the acid-base indicator is bromocresol green, and after inoculation of said medium with a sample containing yeasts of the *Dekkera* and *Brettanomyces* genera, and incubation for 5  
10 to 12 days in adequate conditions for the growth of said yeasts, it is possible to detect the presence, and if needed the enumeration of said yeasts genera, by means of a medium color change, from blue to yellow, and development of cream colored colonies and a phenol-like aroma, characteristic of the yeasts of the *Dekkera* and *Brettanomyces* genera.

15

14—A process for the detection and/or identification of yeasts of the *Dekkera* and *Brettanomyces* genera according to claim 12, characterized in that it is applied to the detection and enumeration of yeasts of the *Dekkera* and *Brettanomyces* genera in food and beverage products samples. A process for the detection,  
20 identification, and/or enumeration of yeasts of the *Dekkera* and *Brettanomyces* genera, characterized by the use of a differential culture medium according to claims 10 and 11, which after inoculation with a food or beverage sample containing yeasts of the said genera and incubation for 5 to 12 days at  $25 \pm 3^\circ\text{C}$ ,  
25 allows the detection, identification and/or enumeration of the said yeasts by means of a change in the medium color from blue to yellow, development of cream colored colonies, and the formation of a phenol-like aroma, being these three features characteristic of yeasts of the *Dekkera* and *Brettanomyces* genera.

15

15-18— Use of a culture medium according to claim 1 ~~claims 10 and 12~~, for  
30 inclusion in an identification gallery, together with other yeast identification tests.

~~16~~ <sup>16</sup> ~~14~~ — Use of a culture medium according to ~~claim 1~~ claims 10 and 12 in an industry, particularly in the quality and process control in a food and beverage industry.

5 ~~17~~ — Use of a culture medium according to claim 10, for inclusion in an identification gallery, together with other identification tests.

~~18~~ — Use of a culture medium according to claim 11, for inclusion in an identification gallery, together with other yeast identification tests.

10

~~19~~ — Use of a culture medium according to claim 10 in an industry, particularly in the quality and process control in a food and beverage industry.

15

~~20~~ — Use of a culture medium according to claim 11 in an industry, particularly in the quality and process control in a food and beverage industry.